was indifferent to the presence of fatty acids in the BSA.

Acidic Dissociation Constant (pK_a). Spectrophotometric titrations to estimate the pK_a of representative nucleosides followed previously described procedures.[26,27]

Biological Activity. Assays of coronary vasoactivity followed a published procedure.[2,17] Each analogue was tested in two dogs, or, if the estimates of molar potency ratio vs. adenosine (MPR) differed by >20%, the analogue was assayed in additional dogs. The MPRs reported here are the arithmetic mean of two assays or the mean ±SEM of three or more assays.

Acknowledgment. This work was supported by Grants-in-Aid from the Suncoast Chapter, Florida American Heart Association Affiliate, Grants NIH HL-26611 and HL-30391, and by Nelson Research and Development, Inc.

Registry No. 1, 38594-96-6; 2, 38594-97-7; 3, 97905-50-5; 4, 97905-51-6; 5, 97948-60-2; 6, 97905-52-7; 7, 97826-32-9; 8, 97826-33-0; 9, 97905-53-8; 10, 97905-54-9; 11, 97905-55-0; 12, 97905-56-1; 13, 20125-39-7; 14, 97826-34-1; 15, 97826-35-2; 16, 97826-36-3; 17, 30784-93-1; 18, 97826-37-4; 19, 97826-38-5; 20, 97826-39-6; 21, 1867-73-8; 22, 5746-27-0; 23, 97826-40-9; 24, 3414-62-8; 25, 19399-25-8; 26, 50474-52-7; 27, 14357-08-5; 28, 17270-22-3; 29, 23096-10-8; 30, 26293-51-6; 31, 15824-83-6; 32, 17270-23-4; 33, 37676-67-8; 34, 97826-41-0; 35, 97826-42-1; 36, 97826-43-2; 37, 97826-44-3; 38, 97826-45-4; 39, 36397-00-9; 40, 36031-53-5; 41, 97826-46-5; 42, 30562-03-9; 43, 57294-65-2; 44, 97826-47-6; 45, 57294-69-6; 46, 57294-70-9; 47, 97374-48-6; 48, 97374-49-7; 49, 41552-82-3; 50, 36396-99-3; 51, 41552-83-4; 52, 41552-84-5; 53, 97826-48-7; 54, 97826-49-8; 55, 57294-66-3; 56, 97826-50-1; 57, 97826-51-2; 58, 97905-57-2; 59, 97826-52-3; 60, 55582-99-5; 61, 97826-53-4; 62, 15763-12-9; 63, 67005-97-4; 64, 4338-48-1; 65, 35662-04-5; 66, 97826-54-5; 67, 97826-55-6; 68, 97826-56-7; 69, 54712-70-8; 70, 97905-58-3; 71, 20330-18-1; 72, 97826-57-8; 73, 97905-59-4; 74, 97905-60-7; 75, 550-33-4; 76, 5399-87-1; 77, 14675-48-0; 78, 2620-62-4; 79, 97826-58-9; 80, 97877-18-4; 81, 5746-29-2; 82, 97826-59-0; 2',3',5'-tri-O-acetylinosine, 3181-38-2; 6-(methylsulfonyl)-9-(β-D-ribofuranosyl)purine, 53821-41-3; (-)-norpseudoephedrine, 37577-07-4; (+)-norephedrine, 37577-28-9.

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Structure–Activity Relationships among Benextramine-Related Tetraamine Disulfides at Peripheral α-Adrenoreceptors

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Several *N,N''*-(dithiodi-2,1-ethanediyl)bis[*N'*-(arylmethyl)-1,6-hexanediamines] were prepared and evaluated for their blocking activity on postsynaptic α_1-adrenoreceptors in the isolated rat vas deferens. The results were compared with those obtained for benextramine (1). *N,N''*-(Dithiodi-2,1-ethanediyl)bis[*N'*-(pyrrol-2-ylmethyl)-1,6-hexanediamine] (pyrextramine, **29**) was the most potent among the tetraamine disulfides investigated. Thus, it was selected for further pharmacological evaluation to assess its receptor specificity. At a concentration of 10 μM it did not affect the responses elicited by 5-hydroxytryptamine and histamine in guinea pig ileum and by isoproterenol in guinea pig atria and tracheal chain. Furthermore, it was more specific than benextramine (1) toward the muscarinic receptor, being significantly less potent in inhibiting the carbachol-induced response in rat jejunum. These results show that pyrextramine (**29**) is an irreversible α-blocking agent that is more potent and specific than benextramine (1). In conclusion, **29** may be a useful tool in the elucidation and characterization of the peripheral α_1-adrenoreceptor.

Two categories of α-adrenoreceptor inhibitors are known: ligands that inhibit through the formation of a covalent bond with some components of the receptor molecule (irreversible blockade) and those that bind reversibly to the receptor, preventing access of the agonist (competitive or reversible inhibition). Among the irreversible inhibitors, β-haloalkylamines have received much attention in the past in the investigation of the α-adrenoreceptor.[1] However, this class of α-blockers is not specific, as other receptor systems are affected as well. In the late 1970s the new class of tetraamine disulfides was developed, and it was shown to be a useful tool in α-adrenergic pharmacology.[2,3] Tetraamine disulfides, exemplified by benextramine (1, BHC), whose main feature is a cystamine moiety carrying aminoalkyl substituents on the nitrogens, have shed new light on the active-site chemistry of the α-adrenoreceptor. It has been demonstrated that 1 irreversibly blocks α_1-adrenoreceptors in the isolated rabbit aorta,[4,5] dog aorta,[6] rat vas deferens,[4,7] rat anococcygeus muscle,[8,9] cat spleen[5], and rat and rabbit atrium.[10,11] Furthermore, it has been shown that the inhibition is time dependent and that this is consistent with covalent bond formation in the drug–receptor interaction, which is the result of a protein thiol–disulfide interchange reaction.[4,12,13] An extensive structure–activity

Chart I. General Structure of Tetraamine Disulfides (Table I)

$$[RNH(CH_2)_nNH(CH_2)_2S-]_2 \cdot 4HX$$

1 (benextramine, BHC): R = 2-MeO-benzyl, $n = 6$

Scheme I

$$[H_2N(CH_2)_6NH(CH_2)_2S-]_2 \xrightarrow{ArCHO,\ NaBH_4} \text{3–39 (Table I)}$$

2

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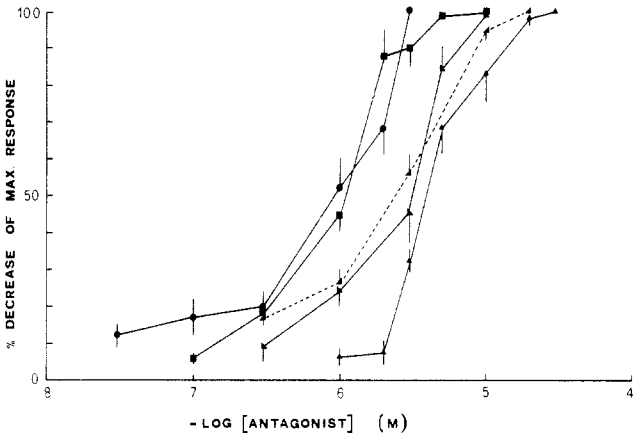

Figure 1. Rat vas deferens; course of α-adrenoreceptor covalent occupancy by **1** (▲), **25** (◢), **26** (◣), **29** (●), and **30** (■). The percent decrease of maximum response to NE was measured after a 30-min incubation for each concentration of antagonist followed by washing with the bath solution for 30 min. The results are expressed as the mean ±SEM of six to eight independent observations.

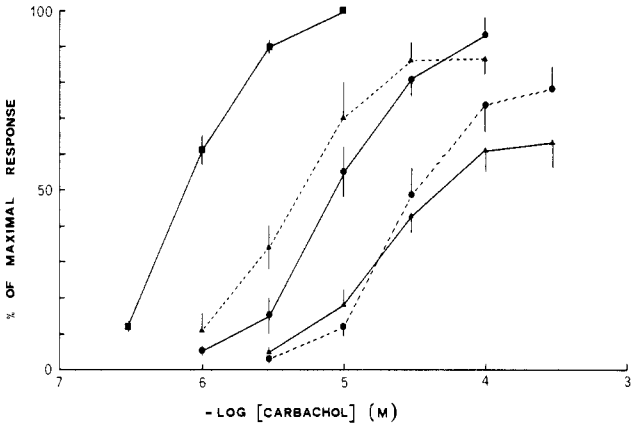

Figure 2. Rat jejunum; cumulative log concentration–response curves to carbachol: of control (■); after exposure to **1** at 10 μM (▲---▲) and 50 μM (▲—▲), and **29** at 50 μM (●—●) and 100 μM (●---●) for 30 min. The results are presented as the mean ±SEM of five to six independent observations.

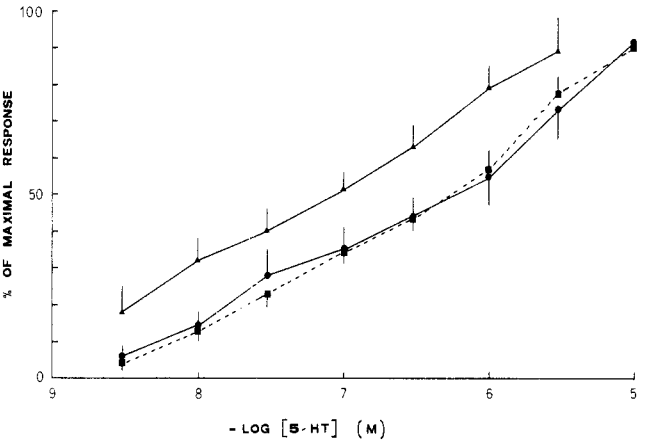

Figure 3. Guinea pig ileum; cumulative log concentration–response curves to 5-HT: of control (■); after exposure to **1** (▲) and **29** (●) at 10 μM for 30 min. The results are presented as the mean ±SEM of six independent observations. The dose–response curve in the presence of **1** was significantly different from control ($p < 0.01$).

relationship study on tetraamine disulfides has shown that optimum α-blocking activity is associated with two different carbon chain lengths separating the inner from the outer nitrogens and depends on the type of substituents on the terminal nitrogens. Thus, optimum α-blocking activity in the unsubstituted series is associated with an eight-carbon chain on rat vas deferens[4] and a seven-carbon chain on rabbit aorta[14] (Chart I; R = H, n = 7, 8). On the other hand, optimum activity in the series with benzyl-type substituents on the terminal nitrogens is associated with a six-carbon chain, as in **1**, on rat vas deferens[4] and rabbit aorta[14] and left atrium.[11] Furthermore, it has been shown that among benzyl-type substituents the 2-methoxybenzyl function of **1** plays a key role in α-blocking activity.

In order to further investigate the effect of substituents on α-blocking activity, we prepared a series of derivatives carrying benzyl-type substituents as well as heteroaromatic groups on the terminal nitrogens of tetraamine disulfides related to **1**.

Chemistry. The structures of the compounds used in the present study are given in Table I and schematically shown in Chart I. They were synthetized according to Scheme I. The substituents on the terminal nitrogens were easily introduced by condensation of **2**[4] with the appropriate aromatic aldehyde and subsequent reduction of the intermediate Schiff bases. Thus tetrahydrochlorides **3–18**, **20**, **22–28**, and **31–37** and tetraoxalates **29** and **30** were obtained in 60–70% yields. The dihydroxybenzyl tetraamine disulfides **19** and **21** were obtained in 50% yield by hydrolysis of the corresponding dimethoxy analogues **18** and **20** with 48% HBr.

Pharmacology. The biological profile of the compounds listed in Table I at α_1-adrenoreceptors was assessed by antagonism of (−)-norepinephrine (NE)-induced contractions of the epididymal portion of isolated rat vas deferens. In order to allow comparison of the results, we used the same techniques and statistical evaluation of the bioassays as for other tetraamine disulfides.[4] Thus, the noncompetitive (irreversible) α-antagonism was determined at a concentration of 20 μM after a 30-min incubation followed by 30 min of washing. The decrease in maximum response was expressed as a percent of the control value. The percent of α-blockade for each compound is expressed as the mean plus or minus SEM of inhibition of the maximum response to 100 μM NE (Table I). Complete concentration–inhibition curves were obtained for the most active tetraamine disulfides **25**, **26**, **29**, and **30** in comparison with that of **1** on rat vas deferens (Figure 1). The most active member of the series, pyrextramine (**29**), was selected for further pharmacological studies,[15] and the results were compared with those obtained for benextramine (**1**). Thus, cholinergic (muscarinic) blocking activity was assessed on rat jejunum by using carbachol as agonist (Figure 2). Furthermore, receptor specificity was studied also by investigating the responses elicited by 5-hydroxytryptamine (5-HT) (Figure 3) and histamine (Figure 4) on guinea pig ileum and by isoproterenol on guinea pig atria (Figure 5) and tracheal chain (results not shown).

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Table I. α-Adrenergic Blocking Results

$[RCH_2NH(CH_2)_6NH(CH_2)_2S-]_2 \cdot 4HX$

compd	R	mp,[a] °C	purifn solvent[b]	formula	anal.[c]	% α-blockade[d]
1	2-MeO-Ph					100 (7)
3	2-F-Ph	272–273	A–B	$C_{30}H_{48}F_2N_4S_2 \cdot 4HCl$	C, H, N	55 (4)
4	3-F-Ph	298–300	A–B	$C_{30}H_{48}F_2N_4S_2 \cdot 4HCl$	C, H, N	73 (4)
5	4-F-Ph	305–306	A–B	$C_{30}H_{48}F_2N_4S_2 \cdot 4HCl$	C, H, N	100 (6)[e]
6	2-NO_2-Ph	232–235	B–C	$C_{30}H_{48}N_6O_4S_2 \cdot 4HCl$	C, H, N	51 (4)
7	3-NO_2-Ph	250–251	A–B	$C_{30}H_{48}N_6O_4S_2 \cdot 4HCl$	C, H, N	49 (4)
8	4-NO_2-Ph	272–273	A–B	$C_{30}H_{48}N_6O_4S_2 \cdot 4HCl$	C, H, N	41 (4)
9	2-Br-Ph	235–236	B–C	$C_{30}H_{48}Br_2N_4S_2 \cdot 4HCl$	C, H, N	58 (4)
10	3-Br-Ph	271–272	A–B	$C_{30}H_{48}Br_2N_4S_2 \cdot 4HCl$	C, H, N	58 (4)
11	4-Br-Ph	299–300	A–B	$C_{30}H_{48}Br_2N_4S_2 \cdot 4HCl$	C, H, N	34 (4)
12	3-Cl-Ph	280–281	A–B	$C_{30}H_{48}Cl_2N_4S_2 \cdot 4HCl$	C, H, N	46 (4)
13	4-Cl-Ph	295–296	A–B	$C_{30}H_{48}Cl_2N_4S_2 \cdot 4HCl$	C, H, N	50 (4)
14	2-MeS-Ph	239–240	B–C	$C_{32}H_{54}N_4S_4 \cdot 4HCl$	C, H, N	66 (3)
15	2-$MeSO_2$-Ph	248–249	B–C	$C_{32}H_{54}N_4O_4S_4 \cdot 4HCl \cdot H_2O$	C, H, N	35 (4)
16	2-MeO-3-Me-Ph	245–246	B–C	$C_{34}H_{58}N_4O_2S_2 \cdot 4HCl$	C, H, N	59 (4)
17	2-MeO-6-NO_2-Ph	214–215	B–C	$C_{32}H_{52}N_6O_6S_2 \cdot 4HCl$	C, H, N	42 (4)
18	3,4-$(MeO)_2$-6-NO_2-Ph	234–235	B–C	$C_{34}H_{56}N_6O_8S_2 \cdot 4HCl$	C, H, N	47 (4)
19	3,4-$(HO)_2$-6-NO_2-Ph	251–252	B–C	$C_{30}H_{48}N_6O_8S_2 \cdot 4HBr$	C, H	99 (5)
20	2-NO_2-3,4-$(MeO)_2$-Ph	210–213	B–C	$C_{34}H_{56}N_6O_8S_2 \cdot 4HCl$	C, H, N	27 (4)
21	2-NO_2-3,4-$(HO)_2$-Ph	220–221	B–C	$C_{30}H_{48}N_6O_8S_2 \cdot 4HBr$	C, H	17 (6)
22	2,4-Cl_2-Ph	271–272	A–B	$C_{30}H_{46}Cl_4N_4S_2 \cdot 4HCl$	C, H	54 (4)
23	3,4-Cl_2-Ph	270–272	A–B	$C_{30}H_{46}Cl_4N_4S_2 \cdot 4HCl$	C, H	39 (4)
24	3,5-Cl_2-Ph	274–275	A–B	$C_{30}H_{46}Cl_4N_4S_2 \cdot 4HCl$	C, H, N	44 (6)
25	2-(5-Me-Furyl)	235–239	B–C	$C_{28}H_{50}N_4O_2S_2 \cdot 4HCl$	C, H, N	100 (5)[f]
26	2-furyl	274–275	B–C	$C_{26}H_{46}N_4O_2S_2 \cdot 4HCl$	C, H, N	100 (5)[f]
27	2-thienyl	280–281	B–C	$C_{26}H_{46}N_4S_4 \cdot 4HCl$	C, H, N	80 (4)
28	3-thienyl	236–241	B–C	$C_{26}H_{46}N_4S_4 \cdot 4HCl$	C, H, N	17 (4)
29	2-pyrrolyl	indef	A–B	$C_{28}H_{48}N_6S_2 \cdot 4H_2C_2O_4$[g]	C, H, N	100 (5)[f]
30	2-(*N*-Me-pyrrolyl)	indef	A–B	$C_{28}H_{52}N_6S_2 \cdot 4H_2C_2O_4$[g]	C, H, N	100 (4)[f]
31	2-pyridyl	245–246	B–C	$C_{28}H_{48}N_6S_2 \cdot 4HCl$	C, H	78 (4)
32	3-pyridyl	251–253	B–C	$C_{28}H_{48}N_6S_2 \cdot 4HCl$	C, H, N	13 (4)
33	4-pyridyl	228–230	B–C	$C_{28}H_{48}N_6S_2 \cdot 4HCl$	C, H	11 (4)
34	3-quinolyl	269–270	B–C	$C_{36}H_{52}N_6S_2 \cdot 4HCl$	C, H, N	36 (4)
35	4-quinolyl	196–201	B–C	$C_{36}H_{52}N_6S_2 \cdot 4HCl$	C, H, N	32 (4)
36	2-naphthyl	291–293	B–C	$C_{38}H_{54}N_4S_2 \cdot 4HCl$	C, H	27 (4)
37	1-naphthyl	293–296	B–C	$C_{38}H_{54}N_4S_2 \cdot 4HCl$	C, H, N	22 (4)

[a] All compounds were recrystallized at least three times and decolorized with charcoal when necessary. Their purity was uniformly checked by TLC (silica) using concentrated NH_4OH–MeOH (6:94) and/or $CHCl_3$–MeOH–HCO_2H (6.5:3:0.5) as mobile phases. Compound **29** was purified as the free base by column chromatography, eluting with concentrated NH_4OH–MeOH (6:94), prior to its transformation into the tetraoxalate salt and subsequent recrystallization. NMR spectra (D_2O) of all compounds were in agreement with the expected structures. The heating rate was 1 °C/min for melting point determinations. [b] A, water; B, methanol; C, 2-propanol. [c] Analyses for the elements indicated were within ±0.4% of the theoretical values. [d] Potencies as irreversible inhibitors of NE-induced responses in rat vas deferens using compound **1** as standard. The number of experiments is in parentheses and the percent inhibition is accurate to within ±6%. For compounds **25**, **26**, **29**, **30**, and **1** complete saturation curves are given in Figure 2. See ref 4 and 15 for detailed procedures. [e] At low concentrations (<20 μM), compound **5** is less active than **1**. [f] At low concentrations (<20 μM), compounds **25**, **26**, **29**, and **30** were more active than **1** as shown in Figure 2. [g] **29** and **30** have indefinite meltings points. They become dark at 190 and 170 °C, respectively.

Results and Discussion

The results assembled in Table I show that all the compounds studied at a concentration of 20 μM have a significant and irreversible α-blocking activity. As regards the substitution of the benzylic moiety of **1** with a heteroaromatic group, the most interesting results were obtained with five-membered rings. Among these substituents the pyrrole nucleus gave both a qualitative and quantitative improvement of the α-adrenoreceptor blocking profile of **1** as outlined below.

Among the derivatives studied at 20 μM concentration, **5**, **25**, **26**, **29** and **30** gave complete inhibition of the α-adrenoreceptor-mediated contraction in the rat vas deferens. Thus, they were studied at lower concentrations, and complete concentration inhibition curves were obtained to compare their potency with that of benextramine (**1**), which was taken as the standard (Figure 1). Compounds **25**, **26**, **29**, and **30** were more potent than **1**, whereas **5** (not shown) was a weaker antagonist. Furthermore, it is also clear that the high activity displayed by **29**, as a consequence of the substitution of the 2-methoxybenzyl group of **1** with a 2-pyrrolyl moiety, represents the most striking result of the present investigation. In fact, **29** caused complete α-adrenoreceptor blockade at a concentration of 3 μM, which is 10 times lower than that of **1**.

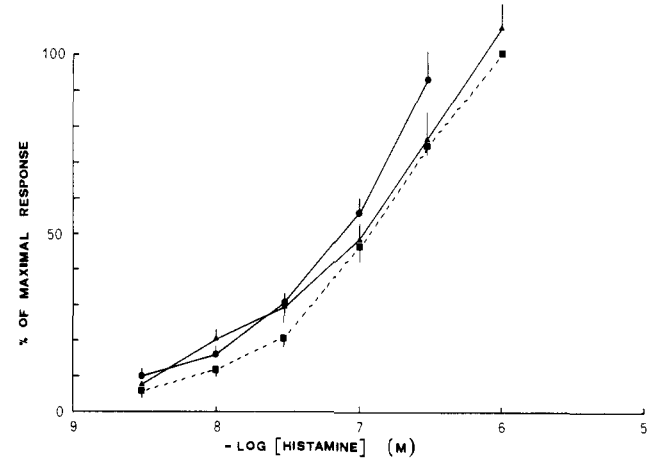

Figure 4. Guinea pig ileum; cumulative log concentration–response curves to histamine: of control (■); after exposure to **1** (▲) and **29** (●) at 10 μM for 30 min. The results are presented as the mean ±SEM of six independent observations.

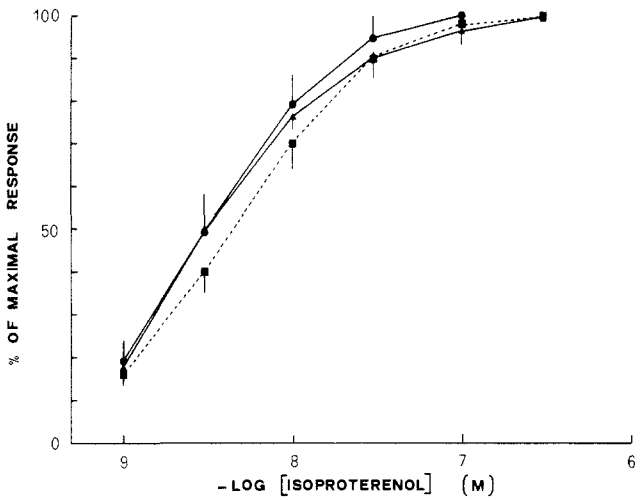

Figure 5. Guinea pig atria; cumulative log concentration–response curves to isoproterenol: of control (■); after exposure to 1 (▲) and 29 (●) at 10 μM for 30 min. The results are presented as the mean ±SEM of five independent observations.

However, at the level of 50% inhibition the difference in activity between **29** and **1** was somewhat less, as indicated by IC_{50} values of 0.90 ± 1.12 and 4.02 ± 0.36 μM, respectively. Pyrextramine (**29**) covalently and irreversibly blocked the α-adrenoreceptor as judged by the observation that inhibition was time dependent and the response was only partially recovered after 5-h washing.[15] In addition to having a similar mechanism of action, pyrextramine (**29**) retains the high receptor specificity outlined for benextramine (**1**). In fact, **29** at a concentration well above that required for complete rat vas deferens α-adrenoreceptor inactivation did not affect the tissue responses elicited by 5-HT and histamine in guinea pig ileum (Figures 3 and 4) and by isoproterenol in guinea pig atria (Figure 5) and tracheal chain (results not shown). In addition, **29** was more specific, as far as antimuscarinic activity is concerned, being less active than **1** in inhibiting carbachol-induced responses in rat jejunum with K_b values of 5.25 ± 0.63 and 2.01 ± 0.80 μM, respectively. Furthermore, **29** and **1** behaved differently as shown in Figure 2. In fact, **29** up to a concentration of 100 μM, caused a parallel shift to the right of the dose–response curve whereas **1**, even at 50 μM, gave a nonparallel shift with depression of the maximum. In contrast to the irreversible α-blockade, inhibition of the muscarinic receptor appeared to be reversible, as shown by the dose ratio values of **29** before and after 30-min washing; these values were 12.66 ± 2.03 and 6.01 ± 1.05, respectively, indicating about 50% recovery.

Besides the significant improvement of both activity and receptor specificity displayed by pyrextramine (**29**) as compared to benextramine (**1**), the present results might give useful information on the mode and site of action of tetraamine disulfides. It is apparent that none of the benzyl-type substituents investigated so far improved the fit with the α-adrenoreceptor compared to the 2-methoxybenzyl function of **1**, which is in agreement with the trend already noted in a previous investigation.[4] However, in light of the results reported for fluoronorepinephrines,[16,17] the biological profile displayed by nitrodihydroxybenzyl tetraamine disulfides **19** and **21** might be relevant. It was shown that the insertion of fluorine into the catechol nucleus of NE gave an effect on biological activity and on specificity toward α- and β-adrenoreceptors that depended on the position of the substitution. Thus, 6-fluoronorepinephrine was an α-adrenergic agonist with virtually no β-agonist activity whereas the 2-fluoro isomer was a β-adrenergic agonist with weak α-activity.[16,17] A possible explanation of this difference in behavior was the alteration of pK_a values and lipophilicity of phenolic groups. Since it has been proposed that the 3,4-dihydroxybenzyl tetraamine disulfide (Chart I: R = 3,4-dihydroxybenzyl, n = 6) might recognize the very same catecholamines binding sites,[2,3] it is not surprising that the nitro group in **19** and **21** had an effect on α-blocking activity, which depends on the position of substitution in the phenyl ring and follows the same trend for NE substitution with fluorine. In fact, the tetraamine disulfide carrying a 3,4-dihydroxy-6-nitrobenzyl substituent (**19**) did not exhibit an α-blocking activity different from that of **1** and the dihydroxy analogue[4] (Chart I; R = 3,4-dihydroxybenzyl, n = 6) whereas the 2-nitro isomer (**21**) was almost inactive at the same concentration of 20 μM (Table I). On the other hand, the insertion of the nitro group at position 6 of the phenyl ring of **1**, giving 6-nitrobenextramine (**17**), resulted in a dramatic decrease in activity. This finding is in agreement with the previous hypothesis that the 2-methoxybenzyl and the 3,4-dihydroxybenzyl groups bind at different sites, which are connected with the same anionic site on the receptor surface.[2,3]

The special role of the methoxy function of **1** has been noted.[4] In fact, it appears essential for optimum α-blocking activity in tetraamine disulfides bearing benzyl-type substituents. Furthermore, the presence of this functionality is responsible for the unusual steepness of the curves relating α-blockade to the concentration of **1**. This may indicate that the 2-methoxy groups are involved in the stabilization of a special receptor conformation displaying some kind of cooperativity.[4] Present results confirm and extend that observation.[15] In fact, all the other benzyl-type substituents investigated did not improve the activity of **1**. However, it is interesting to note that the significantly lower activity associated with **14** might indicate that the methoxy group of **1** plays a role in the receptor binding but not through a dipole–dipole interaction since the sulfur would give an improved interaction owing to the high polarizability of its electrons.

N-methylation of **29**, giving the analogue **30**, does not significantly alter the activity (Figure 1). This may indicate either that the nitrogen of the pyrrole ring is not involved in receptor binding or that it interacts with a site that can accept small substituents such as a methyl group and that the N-proton is not involved in hydrogen-bond formation in the interaction with the receptor molecule. The latter hypothesis, however, appears to be favored since **25–27**, **30**, and **31** having a ring heteroatom oriented as in **29** retain significant activities whereas **28** and **32–35** bearing a differently oriented heteroatom display much lesser activities. Examination of stereomodels of **1** and **29** (as well as **26**, **27**, and **31**) revealed that the aromatic and heteroaromatic rings are superimposable and that the ring heteroatom corresponds to the carbon at either position 2 or position 6 in the phenyl ring, owing to the relatively free rotation about the C–C bond between the ring and the methylene ("benzylic") carbon. Although it is not possible to distinguish between these two possibilities at present, the fact that **29** has the lateral chain α to the pyrrole nitrogen and that in tetraamine disulfides bearing benzyl-type substituents optimum activity is associated

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with ortho substitution might indicate that a $\pi-\pi$ interaction between the aromatic moieties of 1 and **29** and a receptor electron-rich component requires increased electron density on the aromatic carbon bearing the tetraamine disulfide backbone. It is well-known that electron density in the pyrrole system is higher at C2 carbons whereas in substituted methoxybenzenes it is higher at positions 2 and 4, owing to the resonance hybrid of different canonical structures.

Experimental Section

Chemistry. Melting points were taken in closed glass capillary tubes on a Buchi SMP-20 apparatus and are uncorrected. IR and NMR spectra were recorded on Perkin-Elmer 297 and Varian EM-390 instruments, respectively. Although the IR and NMR spectra data are not included (because of the lack of unusual features), they were obtained for all compounds reported and were all consistent with the assigned structures. The microanalyses were performed by the Microanalytical Laboratory of our department, and the elemental compositions of the compounds agreed to within ±0.4% of the calculated value.

Aldehydes. They were obtained commercially with the exception of 2-methoxy-6-nitrobenzaldehyde,[18] 2-methoxy-3-methylbenzaldehyde,[19] 2-(methylthio)benzaldehyde,[20] 2-(methylthio)benzaldehyde *S,S*-dioxide,[21] and 3,4-dimethoxy-2-nitrobenzaldehyde.[22]

***N,N''*-(Dithiodi-2,1-ethanediyl)bis[*N'*-(arylmethyl)-1,6-hexanediamine][23] Tetrahydrochlorides (3–18, 20, 22–28, 31–37) and Tetraoxalates (29, 30).** These compounds were synthesized starting from **2**[4] and the appropriate aryl aldehyde following the procedure described for benextramine (1) and analogues.[4] Yields were in the range of 60–70% (Table I).

***N,N''*-(Dithiodi-2,1-ethanediyl)bis[*N'*-[(3,4-dihydroxy-6-nitrophenyl)methyl]-1,6-hexanediamine][23] Tetrahydrobromide (19) and *N,N''*-(Dithiodi-2,1-ethanediyl)bis[*N'*-[(3,4-dihydroxy-2-nitrophenyl)methyl]-1,6-hexanediamine][23] Tetrahydrobromide (21).** Compound **18** or **20** as the free base (1.0 mmol) was heated under reflux in 48% HBr (10 mL) for 4 h under nitrogen. The solvent was evaporated in vacuo, and the pink-brown residue was recrystallized to give **19** or **21**, respectively, in 50% yield (Table I).

Pharmacology. General Considerations. Tissues from either rats (125–150 g) or guinea pigs (200–300 g) were suspended in 10-mL organ baths containing physiological salt solution (PSS) kept at 37 °C (31 °C for atrial preparation) and aerated with 5% CO_2–95% O_2. The composition of PSS was as follows (mM): 118, NaCl; 4.7, KCl; 2.52, $CaCl_2$; 3.57, $MgCl_2$; 1.54, KH_2PO_4; 12.5, $NaHCO_3$; 11.1, glucose. Dose–response curves were constructed by the method of stepwise cumulative addition of the agonist. The concentration of agonist in the muscle chamber was increased approximately 3-fold at each step, with each addition being made only after the response to the previous addition had attained a maximal level and remained steady. Contractions were recorded by means of a force transducer connected to a two-channel Gemini polygraph.

Rat Vas Deferens and Rat Jejunum. All compounds listed in Table I were tested for α-adrenoreceptor blocking activity in the epididymal portion of the isolated rat vas deferens at a concentration of 20 μM following the procedure reported in ref 4 and 15.

Muscarinic blocking activity of 1 and **29** was evaluated in rat jejunum. The procedure was as given in ref 15.

Guinea Pig Ileum. Segments of guinea pig ileum were used to determine the effects on the histamine- and 5-HT-induced responses by 1 and **29**. Tissues were placed in the muscle chamber under a loading tension of 1.0 g. Submaximal doses (two or three) of agonist were administered 30 min apart to stabilize responses. After this, cumulative dose–response curves were obtained at 20-min intervals with the appropriate agonist until a constant response was obtained. Following a resting period of about 15 min, the antagonist was added and the tissue was again exposed to the agonist subsequent to an incubation period of 30 min.

Guinea Pig Atria. Driven left atrium was used for estimation of β_1-adrenoreceptor blocking activity of 1 and **29**. The preparation was perfused with PSS at 31 °C, loaded with 1.0 g, and stimulated electrically at a frequency of 1 Hz (1-ms duration) with voltages slightly above threshold. The tissue was allowed to equilibrate for at least 1 h before being challenged with isoproterenol, which was the reference agonist. After 30 min of washing and subsequent incubation (30 min) with the antagonist, a second dose–response curve was constructed.

Guinea Pig Trachea. Tracheal chain was used to evaluate β_2-adrenoreceptor blocking activity of 1 and **29**. The preparation was mounted in the muscle chamber, and a tension of 1 g was applied. After a 30-min resting period, a constant level of contraction was obtained by adding carbachol at a concentration of 0.5 μM. As in the case of left atrium, isoproterenol was the agonist and the incubation time for the antagonist under study was 30 min.

Statistical Evaluation. Student's t-test was used to assess the significance of the experimental results.

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Registry No. 1, 68535-69-3; 1 (free base), 69790-18-7; **2**, 68536-04-9; **3**, 97783-07-8; **3** (free base), 97806-85-4; **4**, 97783-08-9; **4** (free base), 97783-42-1; **5**, 97783-09-0; **5** (free base), 97783-43-2; **6**, 97783-10-3; **6** (free base), 97783-44-3; **7**, 97783-11-4; **7** (free base), 97783-45-4; **8**, 97783-12-5; **8** (free base), 97783-46-5; **9**, 97783-13-6; **9** (free base), 97783-47-6; **10**, 97783-14-7; **10** (free base), 97783-48-7; **11**, 97783-15-8; **11** (free base), 97783-49-8; **12**, 97783-16-9; **12** (free base), 97783-50-1; **13**, 97783-17-0; **13** (free base), 97783-51-2; **14**, 97806-84-3; **14** (free base), 97783-52-3; **15**, 97783-18-1; **15** (free base), 97783-53-4; **16**, 97783-19-2; **16** (free base), 97783-54-5; **17**, 97783-20-5; **17** (free base), 97783-55-6; **18**, 97783-21-6; **18** (free base), 97783-56-7; **19**, 97783-22-7; **19** (free base), 97783-57-8; **20**, 97783-23-8; **20** (free base), 97783-58-9; **21**, 97783-24-9; **21** (free base), 97783-59-0; **22**, 97783-25-0; **22** (free base), 97783-60-3; **23**, 97783-26-1; **23** (free base), 97783-61-4; **24**, 97783-27-2; **24** (free base), 97783-62-5; **25**, 97783-28-3; **25** (free base), 97783-63-6; **26**, 97783-29-4; **26** (free base), 97783-64-7; **27**, 97783-30-7; **27** (free base), 97783-65-8; **28**, 97783-31-8; **28** (free base), 97783-66-9; **29**, 97783-32-9; **29** (free base), 92588-09-5; **30**, 97783-34-1; **30** (free base), 97783-33-0; **31**, 97783-35-2; **31** (free base), 97783-67-0; **32**, 97783-36-3; **32** (free base), 97783-68-1; **33**, 97783-37-4; **33** (free base), 97783-69-2; **34**, 97783-38-5; **34** (free base), 97783-70-5; **35**, 97783-39-6; **35** (free base), 97783-71-6; **36**, 97783-40-9; **36** (free base), 97783-72-7; **37**, 97783-41-0; **37** (free base), 97783-73-8; 2-FC_6H_4CHO, 446-52-6; 3-FC_6H_4CHO, 456-48-4; 4-FC_6H_4CHO, 459-57-4; 2-$NO_2C_6H_4CHO$, 552-89-6; 3-$NO_2C_6H_4CHO$, 99-61-6; 4-$NO_2C_6H_4CHO$, 555-16-8; 2-BrC_6H_4CHO, 6630-33-7; 3-BrC_6H_4CHO, 3132-99-8; 4-BrC_6H_4CHO, 1122-91-4; 3-ClC_6H_4CHO, 587-04-2; 4-ClC_6H_4CHO, 104-88-1; 2-$MeSC_6H_4CHO$, 7022-45-9; 2-$MeSO_2C_6H_4CHO$, 5395-89-1; 2-methoxy-3-methylbenzaldehyde, 67639-61-6; 2-methoxy-6-nitrobenzaldehyde, 19689-88-4; 3,4-dimethoxy-6-nitrobenzaldehyde, 20357-25-9; 3,4-dimethoxy-2-nitrobenzaldehyde, 55149-84-3; 2,4-dichlorobenzaldehyde, 874-42-0; 3,4-dichlorobenzaldehyde, 6287-38-3; 3,5-dichlorobenzaldehyde, 10203-08-4; 5-methyl-2-formylfuran, 620-02-0; 2-formylfuran, 98-01-1; 2-formylthiophene, 98-03-3; 3-formylthiophene, 498-62-4; 2-formylpyrrole, 1003-29-8; 2-formyl-*N*-methylpyrrole, 1192-58-1; 2-formylpyridine, 1121-60-4; 3-formylpyridine, 500-22-1; 4-formylpyridine, 872-85-5; 3-formylquinoline, 13669-42-6; 4-formylquinoline, 4363-93-3; 2-formylnaphthalene, 66-99-9; 1-formylnaphthalene, 66-77-3.

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